BACTERIOLOGY OF STOMACH AND DUODENUM IN CASES OF PEPTIC ULCER AND GASTRIC CARCINOMA

BY

MARY BARBER, M.D.

AND

R. H. FRANKLIN, F.R.C.S.

(From the Departments of Surgery and of Pathology, British Postgraduate Medical School)

Unexplained sepsis, which occasionally occurs after apparently straightforward cases of partial gastrectomy, may lead to the development of a subdiaphragmatic abscess or, more rarely, to general peritonitis and death. It has been noted that the tissues surrounding a chronic peptic ulcer are often oedematous, the neighbouring lymph-nodes may be enlarged, and in short the clinical picture may be one of inflammation. The present investigation was undertaken to determine whether any pathogenic bacteria were present in the stomach and duodenum at the time of operation.

The normal bacterial flora of the alimentary tract has been the subject of much work and even more discussion. Billroth (1874) demonstrated that the meconium of newborn infants was sterile and that bacteria appeared with the first yellow stools. Escherich (1885) confirmed this, and showed that the interval elapsing before the appearance of organisms was dependent on when the earliest feed was given. He also established the colon bacillus as the characteristic intestinal organism. Where these bacteria come from, and whether they enter via the mouth or the anus, were for some time subjects of conjecture. In 1900 Cushing and Livingood summed up the evidence then available and concluded that food was the most important source. More recently many studies have been made on the effect of diet on the intestinal flora. Cannon and McNease (1923) found that in rats on a high protein diet the *p*H of the contents of the caecum and colon was 7.0–7.1, and the bacterial flora consisted mainly of proteolytic types, whereas the addition of lactose lowered the *p*H of the intestinal contents and with an increasing acidity the proteolytic bacteria were replaced by aciduric organisms such as *B. acidophilus*. Sanborn (1931a, 1931b) studied the bacteria present in the faeces of adult patients in the Boston Psychopathic Hospital when on various diets. He stated that "a diet high in carbohydrate and one high in meat protein tend to simplify the aerobic flora, respectively, in the acidophilic and the colonic direction." He found, however, that the degree to which this transformation took place varied very much with different individuals, and he concluded that the normal intestinal flora was not simply dependent on age and diet, but was dependent also on the "metabolic characteristics" of the individual.

Action of Bacteria in the Alimentary Tract

Another aspect which has caused considerable controversy is the importance of bacteria in the alimentary tract. Levin (1899) says that Pasteur in 1880 posed the question whether bacteria are essential for digestion. Nuttall and Thierfelder (1895) succeeded in keeping alive and well for eight days a guinea-pig obtained by Caesarean section, confined in a sterile chamber, breathing sterilized air, and fed on sterilized milk. After eight days the animal was killed and the intestines were shown to be free from bacteria. Levin (1899) went to Spitsbergen to study the bacteriology of the Arctic regions. He investigated the intestinal contents of white bears, reindeer, and seals, and found them usually sterile. This he correlated with the great rarity of bacteria in the air and water of the Arctic. The latter contained less than one organism in 11 ml., as opposed to 600,000 per ml. in the Seine. Cohendy (1912) finally proved that animals normally having bacteria in their intestines can survive without them. He reared chicks under sterile conditions for from 12 to 40 days, and found that they developed as well as control chicks, although the food of the former was rather less well digested. For some time after this it was thought that bacteria in the intestinal canal served no useful purpose and were possibly even harmful. In the last few years, however, a new function has been assigned to them—namely, synthesis of vitamins. The ability of many intestinal bacteria to synthesize members of the vitamin B complex *in vitro* is beyond doubt (see Burkholder and McVeigh, 1942). Evidence is now accumulating to show that the biosynthesis of vitamins in the human intestine is by no means negligible. Adolescent males kept on carefully controlled diets deficient in thiamine (Najjar and Holt, 1943) and riboflavin (Najjar *et al.*, 1944) continued in many cases to excrete these substances in normal quantities in the faeces. Ellinger *et al.* (1945) found there was a gross discrepancy between the intake and output of nicotinamide in healthy adults, and that the urinary secretion was reduced by 60%, if the subjects were given succinyl sulphathiazole (sulphasuxidine). From these experiments it cannot be concluded that the intestinal bacteria are necessarily beneficial, as under certain conditions *in vitro* many intestinal bacteria destroy vitamin C (Young, 1942; Young and James, 1942; Young and Rettger, 1943), and organisms of the *Pseudomonas* group will decompose nicotinic acid if that is the only organic compound in the medium (Koser and Baird, 1944). Benesch (1945) obtained a sample of mixed caecal flora from a patient with a caecostomy. He found that under aerobic conditions nicotinic acid was synthesized, and under anaerobic conditions two-thirds of the nicotinic acid present in the original medium was destroyed. Clearly the possible activities of bacteria in the intestines under different conditions are legion and remain a matter for speculation.

Antiseptic Properties of Gastric Juice

In contrast to the prolific flora of the large intestine, the stomach and early duodenum in the healthy adult are usually sterile, except just after a meal. Spallanzani (1783) was the first to draw attention to the antiseptic properties of gastric juice. He demonstrated that "les chairs enfermées avec le suc gastrique ne sont point sujettes à la pourriture." Cushing and Livingood (1900) reviewed a series of cases of gunshot wounds of the abdomen perforating the gut, and found that only those patients survived in whom the lesion was near the pylorus—lesions lower in the alimentary tract leading to peritonitis. This they attributed to the antiseptic action of the gastric juice. Hewetson (1904) gave himself cultures of *Staph. aureus* and *citreus* and *Ps. pyocyanea* via a stomach-tube and then examined samples of gastric juice every 15 minutes. The cocci were found to be killed in 30 to 45 minutes, and the bacilli in 60 to 90 minutes. He noticed no ill effects, and subsequently planted similar cultures in the stomachs of several patients with gastrostomies, obtaining similar results. Knott (1923) correlated the efficiency of gastric juice as a germicidal barrier with the concentration of free hydrochloric acid present, and determined the concentration of the latter necessary to kill various bacteria. He concluded that sporing bacilli and resistant coliforms would easily survive gastric juice; that the typhoid and dysentery group and staphylococci would survive only if the free hydrochloric acid content was low; that *Str. pyogenes, Str. viridans, C. diphtheriae,* and *M. catarrhalis* would pass through to the duodenum only in conditions approaching achlorhydria. Knott and Venables (1924) showed that in 90% of cases with normal gastric acidity the duodenal contents were sterile, whereas Knott (1927) found that 37 patients with pernicious anaemia and achlorhydria all had bacteria in the duodenum. Garrod (1939) showed that susceptibility to the bactericidal action of hydrochloric acid of any pathogenic bacteria entering the body via the alimentary tract varied considerably, and that gastric juice was more bactericidal than hydrochloric acid of equivalent strength.

In gastric disease it has repeatedly been shown that bacteria may actually multiply in the stomach. In 1842 Goodsir described a case "in which a fluid periodically ejected from the stomach contained vegetable organisms of an undescribed form." He described the bacterium in some detail, and suggested for it the name *Sarcina ventriculi*. Gillespie (1893) isolated 24 different organisms from the stomach by means of a stomach-tube. Hewetson (1904) seems to have been the first to study material taken direct from the stomach at operation. He took cultures from the stomach in 36 cases and from the jejunum in 29; 18 of the former and 16 of the latter were sterile. A variety of bacteria were isolated from the rest, but, owing to the fact that they caused no ill effects when injected into rodents, he concluded that they were harmless.

More recent work on the bacteriology of the stomach and duodenum has been carried out mainly to answer two questions: first, whether infection plays a part in the causation of peptic ulcers; and, secondly, whether post-operative sepsis is related to the bacteria present in the stomach at the time of operation. According to Bolton (1913), Böttcher in 1874 demonstrated bacteria in the edges and floor of gastric ulcers and suggested direct infection from the stomach cavity. That bacteria can produce stomach and duodenal ulcers in rodents has been shown by many workers. Some of the earliest results in this line are those of Lettulle (1888), with *Staph. aureus*; Chantemesse and Widal (1888), with a dysentery bacillus; and Charrin and Ruffer (1889), with a filtrate from *Ps. pyocyanea.* Rosenow and Sandford (1915) found streptococci (usually *viridans*) in a very high proportion of peptic ulcers, and Rosenow (1916, 1923) showed that a high percentage of these bacteria, and also cultures of *Str. viridans* isolated from dental infection in ulcer patients, had a "characteristic affinity" for the stomach and duodenum, leading to ulcers in these organs on injection into dogs, monkeys, rabbits, guinea-pigs, and mice, whereas streptococci isolated from other patients did so much less frequently. Similar results were obtained by Christophe (1922), Nakamura (1924), Haden and Bohan (1925), and Nickel and Hufford (1928).

Löhr (1924) was interested in the fact that the free fluid found in the abdomen after perforation of a peptic ulcer was frequently sterile. He studied the contents of the stomach and duodenum obtained at operation from 200 cases; the contents were sterile in 82% of 56 cases of gastric ulcer, in 92% of 40 cases of duodenal ulcer, and in only 78% of 46 cases of gastric carcinoma. Appelmans and Vassiliadis (1932) cultured portions of ulcer obtained at operation from 60 cases. Bacteria were isolated from 13 of 23 duodenal ulcers, 14 of 17 gastric ulcers, and all of 18 gastric carcinomas. The organisms isolated were, in order of frequency, staphylococci, streptococci, yeasts, coliform bacilli, *B. mesentericus,* and lactic acid bacilli. Seley and Colp (1941) investigated the bacteriology of the stomach and duodenum in a series of cases undergoing partial gastrectomy. Actual portions of mucous membrane of these organs were cultured with a view to determining whether post-operative sepsis could in some cases be attributed to bacteria present at the time of operation. Positive cultures were obtained in 93.7% of "gastric malignancies," 83.3% of gastric ulcers, 36.6% of duodenal ulcers, and 37.9% of secondary peptic ulcers. Bacteria regarded as pathogenic, including *Str. haemolyticus, Str. viridans,* non-haemolytic streptococci, *Cl. welchii,* and *Bact. coli,* were isolated from 88% of the "malignancies" and 30% of the benign ulcers. The high percentage of pathogenic organisms in carcinoma cases was correlated with a low average gastric acidity, and the incidence of post-operative complications and mortality appeared higher in these cases. Priestley *et al.* (1944) examined the gastric contents taken at operation from 41 cases. The contents were sterile in 18 of 22 cases of duodenal ulcer, in 1 of 7 cases of gastric ulcer, and in none of 5 carcinomas. Test meals done before operation showed that bacteria were seldom isolated if the level of total gastric acidity was 40 units or higher. The organisms isolated were "green-producing streptococci," micrococci, and Gram-negative bacilli. In spite of this voluminous literature the number of studies where organisms were taken direct from the stomach and duodenum at operation and the bacteria isolated were fully identified is small.

Experimental Tests

In the present study swabs were taken from the mucosa of the stomach and duodenum at operation on a series of 50 patients undergoing partial gastrectomy. Each swab was inoculated on to two blood-agar plates, one of which was then inoculated aerobically and the other anaerobically. Bacteria appearing on these plates were isolated in pure culture and, where possible, identified. In most cases test meals were done before operation and a portion of ulcer was examined histologically.

Table I correlates the gastric condition with the number of positive bacteriological results; in 9 out of 10 cases of gastric carcinoma bacteria were isolated from the stomach alone or stomach and duodenum; the gastric ulcer cases show a lower proportion of positive results, and both swabs were sterile from all of 12 cases of duodenal ulcer.

Table II gives a list of the bacteria grown and the number of times each was isolated in cases of simple ulcer and carcinoma. The last column shows the highest concentration of free hydrochloric acid found in the resting juice, taken before operation, of any patient from whom that particular organism was cultivated. The non-haemolytic streptococci included typical *Str. faecalis* and other types. The coliform bacilli were mostly *Bact. coli,* but *Proteus* and *B. faecalis alcaligenes* were also isolated.

TABLE I

	Simple Ulcers			Gastric Carcinomas	Total
	Gastric Ulcers	Jejunal Ulcers	Duodenal Ulcers		
Stomach and duodenum sterile	12	0	12	1	25
Micro-organisms isolated from stomach; duodenum sterile	11	1	0	6	18
Micro-organisms isolated from stomach and duodenum	4	0	0	3	7
Total	27	1	12	10	50

TABLE II.—*Micro-organisms isolated*

Organism		Simple Ulcers	Gastric Carcinomas	Highest Conc. of Free HCl in Gastric Resting Juice
M. albicans ..	From stomach; 11 cases	9	2	61
Str. viridans ..	,, stomach; 9 ,,	6	3	0
	,, duodenum; 3 ,,	3	0	0
Non-haemolytic streptococci	,, stomach; 7 ,,	2	5	42
	,, duodenum; 2 ,,	0	2	84
Coliform bacilli ..	,, stomach; 7 ,,	1	6	84
	,, duodenum; 3 ,,	1	2	84
Staph. albus ..	,, stomach; 4 ,,	3	1	5
	,, duodenum; 1 case	1	0	0
Neisseriae ..	,, stomach; 4 cases	3	1	0
	,, duodenum; 1 case	1	0	0
Str. pneumoniae ..	,, stomach; 4 cases	2	2	0
Diphtheroid bacilli	,, stomach; 2 ,,	1	1	Not tested
	,, duodenum; 1 case	1	0	,,
Staph. aureus ..	,, stomach; 2 cases	2	0	0
Str. pyogenes (Lancefield Group A) ..	,, stomach; 1 case	0	1	0
	,, duodenum; 1 ,,	0	1	0
Lactobacilli ..	,, stomach; 1 ,,	0	1	0

Discussion

M. albicans, non-haemolytic streptococci, and coliform bacilli were isolated from many patients who had a normal or high gastric acidity. All other bacteria were isolated only from cases where the test meal showed a condition approaching achlorhydria. In this series of 50 cases septic post-operative complications were no higher among the 25 cases with positive cultures than among the other 25. The isolation, however, of such organisms as *Str. pneumoniae, Staph. aureus,* and Lancefield A haemolytic streptococci makes it clear that bacteria in the stomach at the time of operation are a potential source of infection, and pre-operative medication might be advisable at least in those cases with low gastric acidity.

Summary

Swabs for bacteriological examination were taken direct from the mucosa of the stomach and duodenum at operation in 50 cases undergoing partial gastrectomy for peptic ulcer or gastric carcinoma. Bacteria were isolated from 16 of 40 cases of benign ulcer and 9 of 10 cases of carcinoma. Pyogenic cocci (*Str. pneumoniae, Staph. aureus,* and Lancefield A haemolytic streptococci) were isolated from 7 cases.

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TREATMENT OF BRANCHIAL CLEFT CYSTS BY ASPIRATION AND INJECTION OF PURE CARBOLIC ACID

BY

T. FRANCIS JARMAN, M.D.

Deputy Tuberculosis Physician, Welsh National Memorial Association; Deputy Medical Superintendent, Cymla Tuberculosis Hospital, Neath

The occurrence of branchial cysts is relatively rare. The most common mistake in diagnosis is to confuse the condition with an abscess resulting from breaking-down tuberculous glands. In a study of 42 branchial cysts and 9 branchial fistulae, in which incorrect diagnosis was made in 42% of cases, Shedden (1931) reports that 12% were diagnosed as tuberculous glands in the neck. Hamilton Bailey (1928) reports 11 cases of branchial cyst in which this mistake was made, and one very interesting case in which tubercle bacilli were found both in the wall of a branchial cyst and in its fluid contents.

Diagnosis

According to Shedden (1931) branchial cysts occupy a constant anatomical position. They lie in relationship to the deep surface of the upper half of the sterno-mastoid, and nearly always protrude around the anterior border of this muscle. Most commonly the centre of the cyst is opposite the greater cornu of the hyoid bone. In the six cases reported here the swellings were hard and tense, giving an impression of induration rather than of being cystic and fluctuant. Bailey (1923) remarks that all branchial cysts which give rise to symptoms are lined with squamous epithelial cells, and that so-called "mucous branchial cysts" lined with columnar epithelium are in reality cystic hygromas. He states, further, that the presence of cholesterol crystals in aspirated fluid is diagnostic of a branchial cyst. Two other factors, noticed when aspiration is performed, appear to me to indicate a branchial cyst rather than a tuberculous abscess: (*a*) that the fluid content is very thin, and (*b*) that the cyst can almost always be completely emptied at the first aspiration.

The cases here reported were diagnosed as tuberculous cervical adenitis, and were referred to me for treatment of that condition. The diagnosis of branchial cyst was subsequently made in each case on the finding of squamous epithelial cells during examination of aspirated fluid at the Central Tuberculosis Laboratory, Cardiff.

Treatment

The treatment generally recognized as being the only satisfactory one is complete excision (Shedden, 1931; Bailey, 1923, 1928; Baumgartner, 1933). Cutler and Zollinger (1933) report several cases of cervical fistulae and pilonidal cysts treated by a modified Carnoy's solution containing absolute alcohol 6 ml., chloroform 3 ml., glacial acetic acid 1 ml., and ferric chloride 1 g. Robitshek (1933) records a case of branchial fistula successfully treated in this way. Five of the six cases reported below were treated by aspiration followed by the injection of from 0.5 to 1 ml. of pure carbolic acid. In one case the cyst disappeared after a single aspiration without the injection of any sclerosing fluid, but recurred 8 months after this aspiration. Aspiration was made with a moderately fine needle, which was held carefully in place to receive the Record syringe containing the carbolic acid. After aspiration, which in every case caused complete flattening of the cystic swelling, the acid was injected with maximum rapidity in order to obtain a spraying effect with the carbolic on the lining of the cyst wall. Firm pressure was then maintained on the site of the emptied cyst for five or ten minutes.

Case Reports

Case 1.—A man aged 31 was admitted with a very large hard mass in the left side of the neck deep to the upper third of the sterno-mastoid; on Feb. 13, 1943, 25 ml. of rather thin yellowish fluid was aspirated and the swelling completely flattened. On Feb. 27 the swelling reappeared, and 5 ml. of fluid was aspirated. The laboratory report (Dr. Ruth Milne) on the aspirated fluid stated: "No acid-fast bacilli found. The presence of many epithelial cells suggests that the pus may be from a simple branchial cyst. The character of the fluid is also suggestive but not absolutely typical of this." March 20: Swelling very large and hard again; 20 ml. of fluid aspirated, and swelling completely flattened again; 0.5 ml. of pure carbolic acid injected. March 27: Swelling now very small. April 10: Swelling hardly palpable. May 1: Swelling completely gone. Seen in January, 1945, and March, 1946, the patient stated that there had been no recurrence.

Case 2.—A woman aged 41 was admitted on April 2, 1943, with a hard tense swelling in the right side of the neck; 10 ml. of brownish fluid aspirated from the cyst and swelling completely flattened. Laboratory report (Dr. Milne): "No tubercle bacilli. Squamous epithelial cells present in large numbers. Findings typical of a branchial cyst." This case was kept as a control and no carbolic acid was injected. The cyst showed no signs of filling up until Dec. 6. The swelling recurred, and by Jan. 10, 1944, had reached the same size as when originally seen. It was aspirated and 12 ml. of brownish fluid withdrawn. This fluid was again reported on as being characteristic of a branchial cyst. This time, however, 1 ml. of pure carbolic acid was injected after aspiration. Three months later only a small firm swelling was present near the anterior border of the sterno-mastoid. Jan. 29, 1945: No palpable swelling present. March 1, 1946: No recurrence.

Case 3.—A man aged 26 was admitted on Aug. 18, 1943, with a large hard tense mass in the upper half of the right side of the neck, deep to the sterno-mastoid: 12 ml. of thin greenish-yellow fluid aspirated from the swelling. Laboratory report on the fluid (Dr. Milne): "No tubercle bacilli present. Large numbers of squamous epithelial cells. Findings typical of branchial cyst." Sept. 4: Swelling up again; 6 ml. of fluid aspirated; 0.5 ml. of pure carbolic injected. Sept. 18: Swelling much smaller, but up again; 4 ml. of fluid aspirated; 1 ml. of pure carbolic acid injected. Dec. 13: Now has a small firm swelling near the anterior border of the sterno-mastoid. Jan. 10, 1944: A tiny hard nodule only is palpable. Jan. 29, 1945: No mass or nodule palpable. April 20, 1945, and March 1, 1946: No recurrence.

Case 4.—A young woman aged 22 was admitted on March 3, 1944, with a tensely fluctuant swelling in the upper half of the left side of the neck; 16 ml. of brownish thin fluid aspirated. Laboratory report on fluid (Dr. Milne): "No tubercle bacilli present. Large numbers of squamous epithelial cells observed. Findings typical of branchial cyst." April 28: Swelling up again; 18 ml. of fluid aspirated and swelling completely flattened; 1 ml. of pure carbolic injected. May 12, 1944: Small hard mass palpable. July 21: Swelling completely gone; nothing to palpate. Jan. 22, 1945: No palpable swelling. April 23, 1945, and March 1, 1946: Follow-up; no recurrence.

Case 5.—A young woman aged 23 was admitted on March 3, 1944, with a large tensely fluctuant swelling in the left side of the neck; 15 ml. of thin yellowish fluid aspirated from the swelling. Laboratory report on the fluid (Dr. Milne): "No tubercle bacilli; large numbers of squamous epithelial cells. Findings typical of a branchial cyst." March 10: 5 ml. of fluid aspirated; 1 ml. of pure carbolic injected. April 28: Swelling completely gone. May 26: Swelling still absent; nothing at all abnormal to palpate. Jan. 29, 1945: Swelling reappeared a week ago. A very hard swelling the size of a walnut present. Aspiration attempted but no fluid obtained. Diagnosis doubtful. Feb. 10: Swelling has disappeared; may have been a lymphadenitis. April 30: No palpable swelling. March 3, 1946: No recurrence.

Case 6.—A girl aged 12 was admitted on Dec. 22, 1944, with a fairly tense cystic swelling in the left side of the neck, deep to the